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Silica as a Potential Controlling Agent for the Blue-green Alga *Aphanizomenon flos-aquae* (L.) Ralfs.

By

Glen D. Johnson

and

Dean W. Blinn

Prepared for the U.S. Forest Service by:

Department of Biological Sciences
Northern Arizona University
Flagstaff, Arizona 86001

Wildlife Habitat Technical Bulletin No. 5

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Glen D. Johnson
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Dean W. Blinn

Department of Biological Sciences
Northern Arizona University
Flagstaff, Arizona 86011

The research described in this report
was supported by funds provided by
The U.S. Forest Service,
Wildlife Management Branch,
under contract number 102/932.01.29

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The U.S. Forest Service, Region 3 financed this study in order to find a suitable and natural method of influencing and managing algae growths.

ABSTRACT:

Unialgal cultures of Aphanizomenon flos-aquae (L.) Ralfs were subjected to various concentrations of silica in order to determine the potential of silica to reduce populations of this nuisance blue-green alga. The concentration of silica introduced as $Na_2SiO_3 \cdot 9H_2O$, ranged from amounts as low as 1.4 mg l^{-1} to as high as 90 mg l^{-1}. Initial and final pH was recorded as well as final silica concentration, filaments per ml, cells per ml, chlorophyll a and total carotenoids.

Within the 95% confidence interval, silica concentrations up to 45 mg l^{-1} demonstrated no significant inhibition upon A. flos-aquae grown in unialgal cultures. However, concentrations of silica above 50 mg l^{-1} appeared to significantly reduce cell density. The feasibility of treating a lake with enough silica to maintain the concentration at this level is obviously prohibitive. It is proposed, however, that during periods of reduced silica concentration, elevating the silica concentration to the range normally encountered in aquatic systems, may enhance species competition, especially with diatoms. This in turn may reduce the potential for blue-green algal blooms. The interactions of algal species in natural communities have not been studied in this respect and it is proposed that more research be directed along these lines.

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INTRODUCTION:

The use of aquatic herbicides has increased in this country as industrial and agricultural practices continue to enrich our rivers, lakes, and ponds. With this increase in nutrients, particularly nitrates and phosphates, there is a concomitant increase in algal populations. During the warm summer months when these populations tend to bloom, economic as well as ecological problems normally arise. Severe algal blooms may clog city water filters, produce taste and odor problems, result in fish kills, and cause other undesirable changes in the physical and chemical qualities of the water (Courchene and Chapman, 1975; Maddox et al., 1971; Mullison, 1970; Robson, 1970; and Valcik, 1975).

As an algal control, it should not be the intention of any herbicide treatment to completely eliminate all algal populations. For the complete elimination of harmless species might contribute to the increased growth of other more objectionable species (Courchene and Chapman, 1975). As the utilization of herbicides increase there is the realization of the possible existence of a biological magnification problem through herbicidal use (Mullison, 1970). This must be fully understood before any herbicides are applied as it may lead to secondary effects involving the health of domestic animals, wildlife, aquatic organisms, and even man (Maddox et al., 1971). Therefore, more research should be placed on this aspect of the subject in developing new products.

If a natural component of the aquatic environment can be made useful in controlling algal blooms then these secondary effects may be greatly reduced. Unfortunately, research along these lines has been lacking. Examination of the literature reveals that there is a negative relationship between blue-green algal populations and increased silica concentration

(Button and Blinn, 1973, 1975; Cole, 1975; Golterman, 1975; Hutchinson, 1967; Schelske and Stoermer, 1972; and Wetzel, 1975). It is generally observed that as the silica concentrations decrease in late spring, the blue-green algal populations increase. Whether this increase is due directly to the drop in silica concentration or to interactions in species succession or both is not fully understood.

Since silica makes up 50% of the earth's crust and is found in all aquatic systems it is only reasonable to assume that if it has any algicidal properties it may have less detrimental effect upon the environment than complex industrial chemicals. In natural waters the range of silica most commonly observed is from 1-30 mg l^{-1} with the mean for surface water at 13 mg l^{-1} (Hem, 1975). Silica occurs in natural waters as a result of the action of erosion on the rocks and soil. Largest concentrations are found in groundwater associated with volcanic rocks while lowest amounts of silica are found in waters associated with carbonate rocks. The form that silica takes in natural waters is the monomolecular silicic acid, H_4SiO_4. The concentration of silica within lakes frequently exhibits distinct variation in both seasonal and spatial distribution. The intense assimilation of silica by diatoms for cell wall construction during spring and fall periods and the sedimentation of these diatom frustules more rapidly than silica input by surface and groundwater is the most important silica sink (Wetzel, 1975). Temperature and the difference in silica concentration between the sediment and the overlying water influences the rate of release of silica from the sediment back into the aquatic system.

Ashurst Lake, Arizona, has had a history of summer algal blooms. Prior to a herbicide treatment in the summer of 1974, there had been sixteen consecutive years of blue-green algal blooms (Schwartz, 1976). These blooms

ranged in intensity from being unsightly and fowl smelling to causing extensive fish kills. They also occur in relatively cool waters as the maximum water temperature of this mountain lake (2133m) reaches 21°C. Late in the spring of each year the silica content of this lake decreases from a previous high of 9.2 mg l^{-1} to less than 0.5 mg l^{-1} (Schwartz, 1976; Johnson, unpublished data) which corresponds to the increases in blue-green algal populations.

If silica can be maintained at an appreciable amount by the addition of soluble silica to the system, then control of these blue-green algal blooms may be possible. This laboratory study was consequently initiated to explore the quantitative and qualitative effects of silica as a potential algicide on the nuisance blue-green alga Aphanizomenon flos-aquae. A more complete list of references concerning the role of silica in aquatic systems can be found in the Appendix of this report.

MATERIALS AND METHODS:

ALGAL CULTURES: Aphanizomenon flos-aquae (L.) Ralfs was isolated from Ashurst Lake, Arizona, into a biphasic soil-water medium (Pringsheim, 1946) during November, 1976. Stock cultures were maintained in a culture room under growlux illumination at 3200 lux, at a photoperiod of 12:12, and at 20 ± 2°C which is comparable to temperatures in Ashurst Lake during the bloom period (Schwartz, 1976; Johnson, unpublished data).

EXPERIMENTAL TRIALS: In determining the response of elevated silica concentrations on the growth of A. flos-aquae, four separate laboratory trials were performed. The first trial was conducted with duplicates of each silica concentration while the remaining three were conducted with triplicates.

In preparation for each trial, 250 ml Erlenmeyer flasks were sterilized with cotton stoppers intact. Once the flasks had cooled to room temperature, $Na_2SiO_3 \cdot 9H_2O$ (sodium meta-silicate) was added to each flask to provide the

desired concentration of silica. Silica concentrations varied between 1.4 mg l^{-1} to 90 mg l^{-1} depending on the experimental trial. Prepared Carolina Alga-Gro^{R} Freshwater medium (Carolina Biological Supply Co.) and Chu #10 medium (Gerloff, et al., 1950) were added to each flask at a ratio of approximately 1:4 respectively. The Chu #10 medium was modified however, in that the $Na_2SiO_3 \cdot 9H_2O$ constituent was omitted to help maintain a negligible silica concentration in the culture systems. A Hach DR-EL/2 kit was employed to determine the silica concentration in the prepared Carolina medium. Mean silica concentration in this preparation was 0.34 mg l^{-1}. A 1.0 ml inoculum of A. flos-aquae from the stock cultures was added to each flask. The flasks were allowed to incubate under growlux illumination at 3200 lux, at a temperature of approximately 20 ± 2°C, and a photoperiod of 12:12. Cultures were incubated from 12 to 22 days.

Prior to inoculation of A. flos-aquae, 25 ml was removed from each flask to determine the initial pH. This was measured on a Beckman Expand-Mate pH meter. At the termination of each incubation period, solutions were analysed for final pH and silica concentration using the Beckman pH meter and a Hach DR-EL/2 kit respectively. Number of filaments per ml, cells per ml, chlorophyll a and total carotenoids were also measured. A 1.0 ml aliquot was drawn off initially and because of the high number of filaments present in most flasks, a 1:10 dilution was necessary in order to make accurate counts. A Sedgewick-Rafter counting chamber was used for estimating filaments per ml. Due to the variance in cell number per filament, it was necessary to count cells in the first twenty filaments encountered for each culture to establish an average cell number per filament. This average for cells per filament was used as a multiplication factor to estimate cells per ml. Twenty-five ml was removed from each flask to determine the chlorophyll a and total carotenoids. The extracted pigments were analyzed according to Strickland

and Parsons (1968) on a Model 124 Perkin-Elmer Double Beam Spectrophotometer. All results were recorded and averaged respectively for each incubation period. Computer analysis of the data was performed on a Xerox Sigma VI using the MiniTab program (Ryan, et al., 1976) to determine the one-way and two-way analysis of the variance.

RESULTS AND DISCUSSION:

There were a total of four separate laboratory trials or incubation periods performed to determine the effects of elevated silica concentrations on population growth of Aphanizomenon flos-aquae. The first two trials were used only to give a preliminary estimate on silica concentrations to be tested and to establish the duration of the incubation period. By terminating half of each of these two trials after approximately two weeks and allowing the second half to continue for another week, it was determined that maximum population development had occurred by the end of 12 to 14 days. Therefore, the remaining two trials were allowed to incubate for this duration before populations were estimated.

From the results of the first two trials it was discovered that due to cultural aberations, that is age and health of the algal cells at the time of inoculation, it was very difficult to exactly reproduce the results from one trial to the next (Table I and II). The age of the algal cells or more specifically the metabolic condition of the cells at the time of inoculation into the growth medium is extremely important in determining their performance. Whether the cells are in the log or lag phase will be a major factor as to how the cells respond to the silica treatment. For example, young cells may be either more susceptible or less susceptible to the silica treatments than cells in a different metabolic state. For our experiments every effort was made to insure obtaining inoculations of algal cells of approximately

the same stage of development from one trial to the next in order to eliminate this variable. Examination of the data presented in Figures I and II suggest a slightly different metabolic stage for the two stock cultures due to higher cell densities attained in Figure II; however, response to silica is not significantly different for each trial.

Temperature, growlux illumination, and photoperiod all remained the same throughout all flasks and from trial to trial. The possible variables that could effect the performance of the A. flos-aquae cells would be pH and/or silica concentration. As is evident from Table I and II, the initial pH increased as the silica concentration increased. This was the direct response of adding the salts of the $Na_2SiO_3 \cdot 9H_2O$ to each flask. The pH values encountered in the culture systems were not considered extreme since under natural conditions maximum populations of this strain of A. flos-aquae from Ashurst Lake are found at a pH range of 9.0 to 9.7 (Schwartz, 1976). Statistical analysis of the changes in pH from flask to flask within any treatment and between silica treatments indicates that there was no significant change between these levels within the 95% confidence interval. Therefore, the changes in pH in this study due to the silica treatments appear to have a negligible influence on the growth of A. flos-aquae, suggesting that the main variable in the culture systems is silica concentration.

With an increase in silica concentration up to approximately 35 mg l^{-1}, there appears to be a direct increase in cell density. Higher silica concentrations appeared to decrease cellular division and growth. This apparent trend is depicted in both incubation periods presented in Figures I and II. Figure I shows an increase in cells per ml from 1.08×10^6 to 1.64×10^6 as the silica concentration increased from 1.9 to 27.9 mg l^{-1}. Then at 38.3 mg l^{-1} silica, the cells per ml decreased to 0.9×10^6 (Table I). This same trend is shown in Figure II as the cells per ml increased from 1.95×10^6

to 2.45×10^6 as the silica concentration increased from 1.4 to 34.1 mg l^{-1}. At 45 mg l^{-1} silica, the cells per ml decreased to 1.49×10^6 (Table II). A statistical approach was therefore undertaken to analyze the data from both trials. A one-way analysis of variance indicated that within the 95% confidence interval there was no significant difference in cell estimates among the various silica treatments. Further scrutiny of these data using the two-way analysis of variance indicated that even within the replications of the same silica concentration there were no significant differences among cell estimates at the 95% confidence level. This means that there is no significant difference in cell estimates either within replications of the same silica concentration or among the various silica treatments. It therefore suggests that silica concentrations up to 45 mg l^{-1} have neither an inhibitory nor stimulatory effect upon A. flos-aquae. However, it was observed in preliminary trials that substantial inhibition of A. flos-aquae had occurred in cultures containing over 50 mg l^{-1} of silica.

The preceeding conclusion can only be applied to laboratory unialgal cultures. Under natural conditions interacting factors, the most important being competition among algal species, may allow silica to be a useful tool in controlling A. flos-aquae. It might be hypothesized that if seasonal periodicity could be altered to give other more desirable species a competitive advantage over A. flos-aquae, then the bloom of this blue-green alga may be significantly reduced.

In temperate lakes such as Ashurst Lake, phytoplankton biomass begins to increase in the spring following ice breakup and improved light and temperature conditions. This spring maximum most commonly consist of diatoms that are tolerant to low temperatures (Wetzel, 1975). The spring maximum is then followed by a slight decrease in phytoplankton biomass which

subsequently leads into a summer profusion of blue-green algae that persist until disruption of thermal stratification. Wetzel (1975) states that seasonal succession of phytoplankton biomass is reasonably constant from year to year unless altered by outside interferences such as man's activities.

The decline of diatom species during the late spring has been shown to be directly correlated to the decrease in silica concentration (Golterman, 1975; Wetzel, 1975). As the diatom biomass increases during the spring maximum the silica content of the water decreases due to its utilization in cell wall formation. It has been shown that silica concentration is the growth yield-limiting factor for diatoms (Golterman, 1975). As the silica-depleted diatom cells die and summer stratification develops, the blue-green algae predominate. Golterman (1975) believes it is the competition for nutrients which is the operative factor under natural conditions that regulates seasonal periodicity.

Theoretically, if the silica content is not depleted diatom cells will continue to grow and reproduce thereby not creating a vacuum for blue-greens to fill. This in essence might be enough to offset the competitiveness of A. flos-aquae and other "pest species" and thereby eliminate the seasonal summer blooms.

Maintaining a soluble silica concentration in the ranges necessary to support a diatom population is very plausible. Golterman (1975) and Wetzel (1975) report silica solubility concentrations as SiO_2 as high as 60-80 mg l^{-1} at 0^oC and 100-140 mg l^{-1} at 25^oC within pH values commonly encountered. Problems may arise however, with possible large increases in pH values when using Na_2SiO_3 forms of silica (Golterman, 1975). How these increases in pH will affect the aquatic system is difficult to predict. in fact, the natural buffering system may absorb the addition of the silica

salts reducing the change in pH substantially. Fortunately, silica in other forms, specifically TES ($Si(OC_2H_5)_4$), does not appear to significantly increase the pH (Hem, 1975).

With the continuing high cost of chemicals, it may be prohibitive to treat large size lakes with enough silica to be effective, however, this needs to be tested experimentally in situ. Unfortunately, mechanical or other chemical means of controlling algal blooms are probably just as expensive to use. Ashurst Lake, Arizona, contained 2200 acre feet of water at the end of the summer of 1977 (Arizona Game and Fish Department, pers. comm.). Depending on the concentration of silica applied it could be relatively expensive to treat this lake at the present rate of $2.65/lb. for commercial grade bulk rate sodium meta-silicate (J.T. Baker Co.). The more important the lake or pond is in terms of its recreation and watershed value the easier it is to justify a costly application of any chemical. The benefits must be weighed carefully before decisions can be made on whether or not to treat. Obviously more data needs to be collected on this subject, particularly from in situ conditions before we can answer all the questions regarding the application of silica as a blue-green algicide.

It is recommended that an experimental bag study containing natural algal communities be conducted to test the effectiveness of silica under natural competitive conditions. This might be performed by suspending large clear plastic bags containing lake water in the water column and adding the silica directly to the contents of the bags. Anchoring the bags would keep them from drifting apart. Algal populations and physico-chemical features could be monitored at regular intervals which would allow accurate monitoring of the effects of the silica treatments.

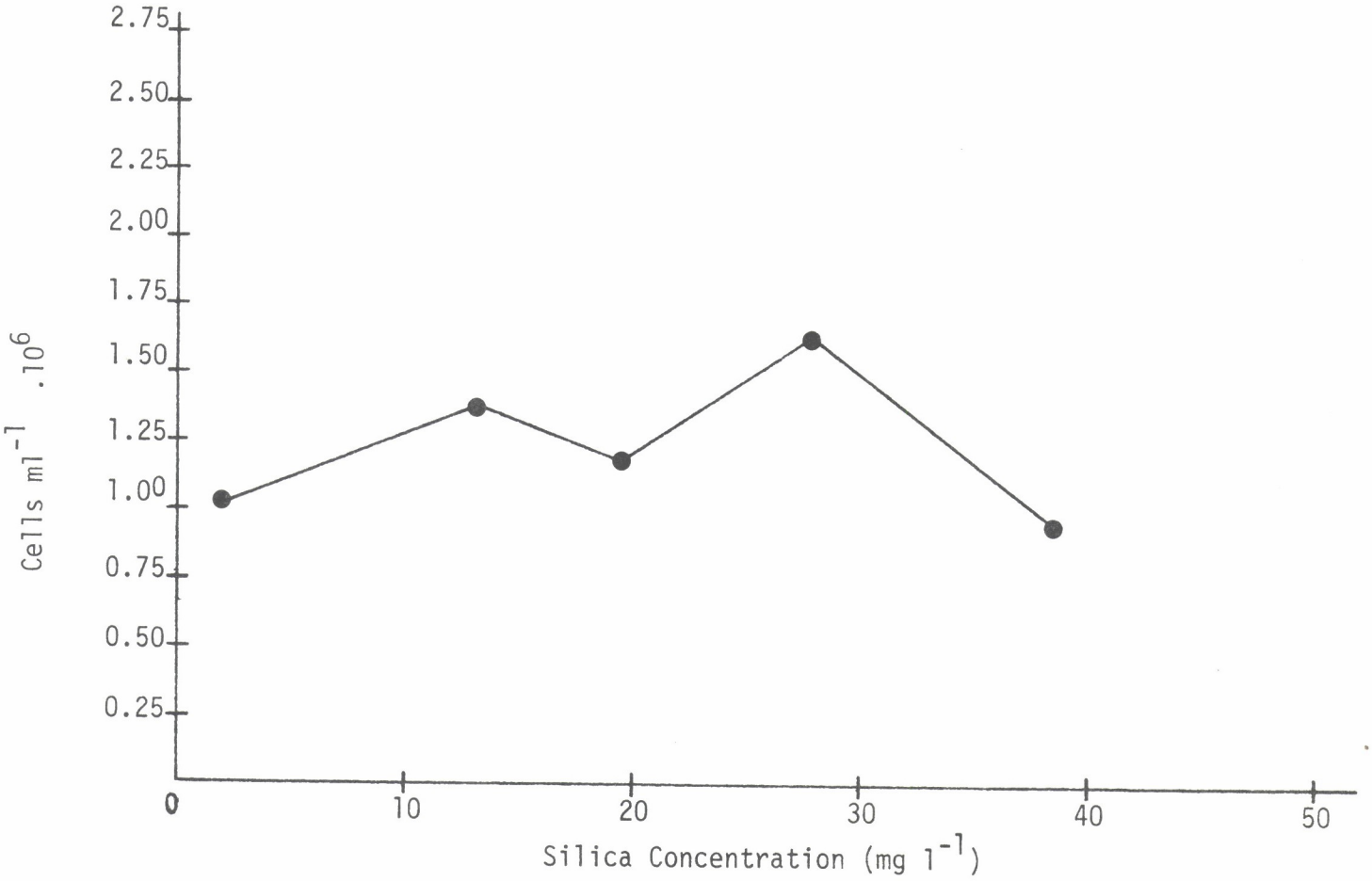

Figure 1. Effects of various silica concentrations on Aphanizomenon flos-aquae for a 12 day incubation period

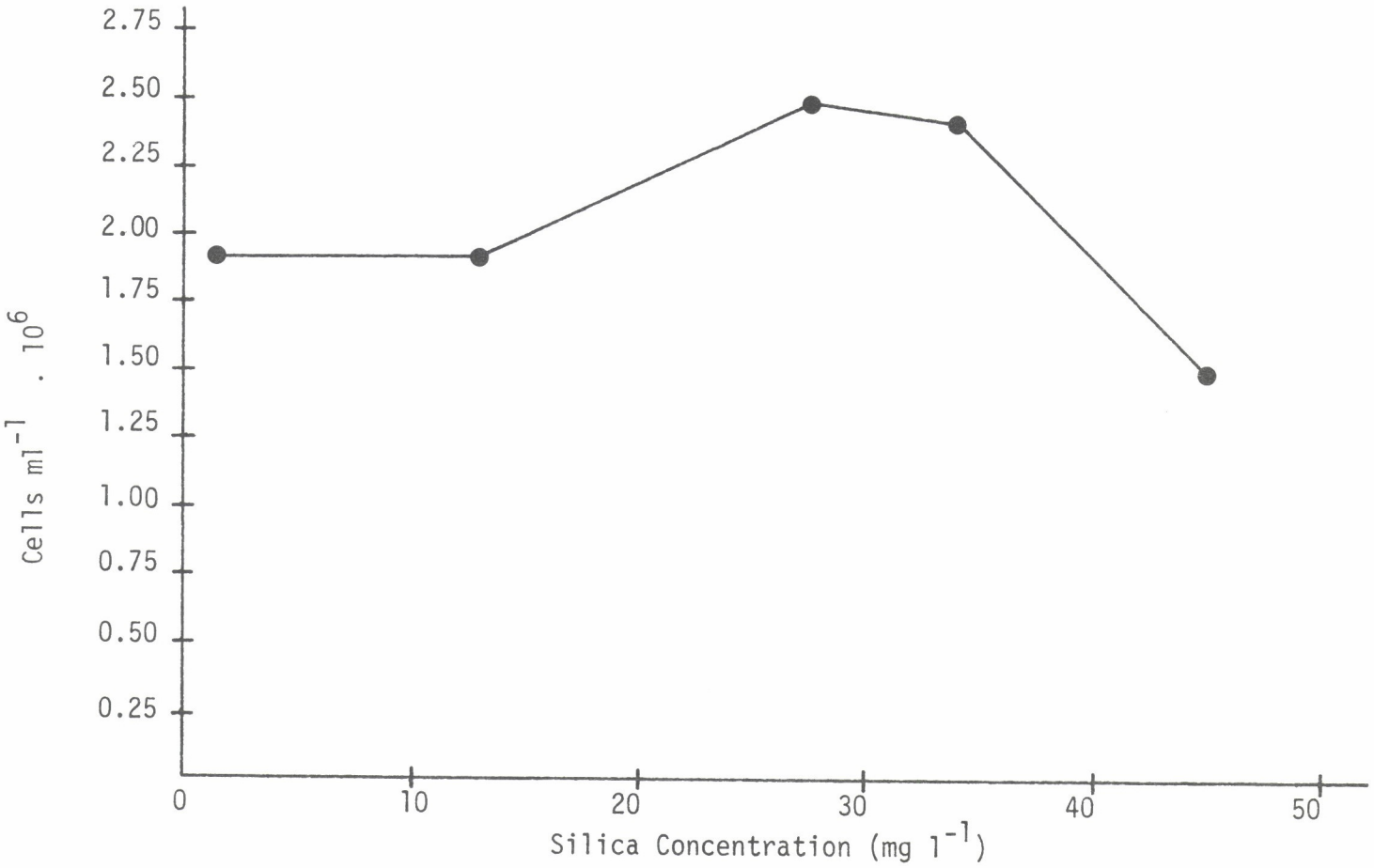

Figure 2. Effects of various silica concentrations on Aphanizomenon flos-aquae for a 13 day incubation period

TABLE I. Effects of various silica concentrations on cell density of Aphanizomenon flos-aquae. All values represent the average of three replications for 12 day incubation period.

Silica Concentration mg l^{-1}	pH*	pH**	Cells ml^{-1}	Chlorophyll a mg m^{-3}	Total Caratenoids mg m^{-3}
1.9	8.3	9.7	1.09×10^6	0.2985	0.1563
13.3	9.7	10.0	1.32×10^6	0.3046	0.1699
19.5	10.0	9.7	1.22×10^6	0.2336	0.1251
27.9	10.2	10.1	1.64×10^6	0.2819	0.1448
38.3	10.4	9.6	0.90×10^6	0.1744	0.0943

*Average pH values for the three replications at the initiation of the trial.

**Average pH values for the three replications at the termination of the trial.

TABLE II. Effects of various silica concentrations on cell density of Aphanizomenon flos-aquae. All values represent the average of three replications for 13 day incubation period.

Silica Concentration mg l^{-1}	pH*	pH**	Cells ml^{-1}
1.4	8.3	9.3	1.95×10^6
12.7	9.4	9.5	1.97×10^6
27.5	9.8	9.9	2.50×10^6
34.1	10.0	10.1	2.45×10^6
45.0	10.1	9.4	1.49×10^6

*Average pH values for the three replications at the initiation of the trial.

**Average ph values for the three replications at the termination of the trial.

SUMMARY:

1) Laboratory tests indicate that silica concentrations as high as 45 mg l^{-1} have no significant effect upon the growth of A. flos-aquae. However, concentrations of silica above 50 mg l^{-1} appear to substantially reduce cell density.

2) It is suggested that maintaining silica concentrations capable of supporting an abundant diatom population (>5ppm) may, through competitive interactions, control or significantly reduce A. flos-aquae cell numbers below bloom conditions. Therefore, the rates of application of silica to a natural body of water to control A. flos-aquae may be in the ranges normally encountered in aquatic ecosystems and still be effective due to competitive interactions of algal species.

3) The addition of silica, a natural component of any aquatic system, should have less detrimental effects upon all the biological life forms associated with an aquatic system than standard industrial chemicals or mechanical means.

4) An in situ study with natural algal communities needs to be conducted to insure that the use of silica as a controlling agent for blue-green algae is ecologically balanced before decisions can be made about its use.

LITERATURE CITED

Button, K.S. and D.W. Blinn. 1973. Preliminary seasonal studies on algae from Upper Lake Mary, Arizona. J. Ariz. Acad. Sci. 8: 80-83.

_____ 1975. Planktonic diatom fluctuations in a northern Arizona mountain lake. Southwest. Nat. 20: 397-408.

Cole, G.A. 1975. Textbook of limnology. The C.V. Mosby Co., St. Louis, Mo.

Courchene, J.E. and J.D. Chapman. 1975. Algae control in Northwest reseservoirs. J. Am. Water Works Assoc. 67: 127-130.

Gerloff, G.C., G.P. Fritzgerald, and F. Skoog. 1950. The isolation, purification, and culture of blue-green algae. Am. J. Bot. 37: 216-218.

Golterman, H.L. 1975. Physiological limnology: Developments in water science. Elsevier Scientific Pub. Co., Amsterdam.

Hem, J.D. 1975. Study and interpretation of the chemical characteristics of natural water. 2nd Ed. Geological Survey Water-Supply Paper 1473.

Hutchinson, G.E. 1967. A treatise on limnology. Introduction to lake biology and the limnoplankton. Vol. 2. John Wiley and Sons, Inc.

Maddox, D.M., R.D. Blackburn, L.A. Andres, N.R. Spencer, R.D. Hennessey. 1971. Insects to control alligator weed - an invader of aquatic ecosystems in the U.S. Bioscience 21: 985-991.

Mullison, W.R. 1970. Effects of herbicides on water and its inhabitants. Weed Sci. 8: 738-750.

Pringsheim, E.G. 1946. The biphasic or soil-water culture method for growing algae and flagellata. J. Ecol. 33: 193-204.

Robson, T.O. 1970. Water weed control: the problem in Britian. Chem. Ind. 45: 1419-1423.

Ryan, T.A., B.L. Joiner, and B.F. Ryan. 1976. MiniTab: Student handbook. Duxbury Press, North Scituate, Mass.

Schelske, C.L., and E.F. Stoermer. 1972. Phosphorus, silica, and eutrophication of Lake Michigan. Pages 157-171 in G.E. Likens, ed. Nutrients and eutrophication: the limiting-nutrient controversy, ASLO Special Symposia 1. American Society of Limnology and Oceanography, Lawrence, Kansas.

Schwartz, S. 1976. Responses of physico-chemical parameters and plankton populations to treatment with aquazine at Ashurst Lake, Arizona. MS Thesis, Northern Arizona Univ.

Strickland, J.D.H., and T.R. Parsons. 1968. A practical handbook of seawater analysis. Fisheries Res. Bd. of Canada, Ottawa.

Valcik, J.D. 1975. Algae in Baltimore's reservoirs. J. Am. Water Works Assoc. 67(3): 109-113.

Wetzel, R.G. 1975. Limnology. W.B. Saunders Co., Philadelphia, Penn.

APPENDIX

LITERATURE REVIEW

The following list is in compliance with the contract concerning a literature review of recent articles on the role of silica in aquatic systems.

Allen, H.L., and B.T. Ocevski. 1976. Limnological studies in a large, deep oligotrophic lake. Arch. Hydrobiol. 77: 1-21.

Balonov, I.M. 1973. The colorless chrysomonas alga Paraphysomonas vestita (Stokes) Saedeler. Mikrobiologiya 41: 563-565.

Bennekon, J.J. van. 1974. The seasonal cycles of reactive silicate and suspended diatoms in the Dutch Wadder Sea. Neth. J. Sea Res. 8:174-207.

Chen, D.M.H., and P.A. Tyler. 1976. Nutrient economies and trophic status of Lake Sorell and Crescent, Tasmania. Aust. J. Mar. Freshwater Res. 27: 151-163.

_____ 1973. The effect of diatom populations on silica concentrations of Lake Sorell and Crescent, Tasmania, and the utilization of tripton as a source of silica. Br. Phyco. J. 8: 249-256.

Chen, H.C. 1971. Increase in benthic algae in milkfish ponds by application of silicates. Fish. Serv. Chin. - Am. Jt. Comm. Rural Reconstr. 11: 71-83.

Codispoti, L.A., and D. Lowman. 1973. A reactive silicate budget for the Arctic Ocean. Limnol. Oceanogr. 18: 448-456.

Conway, E.L., P.J. Harrison, and C.O. Davis. 1976. Marine diatoms grown in chemostats under silicate or ammonium limitations. Mar. Biol. 35: 187-199.

Dickson, E.L. 1975. A silica budget for Loughneagh 1970-1972. Freshwater Biol. 5: 1-12.

Edmond, J.M. 1973. The silica budget of the Antarctic circumpolar current. Nature 241: 391-393.

Goering, J.J. 1974. Silicic acid uptake by marine phytoplankton blooming near a marine sewage outfall located in the Gulf of Saronikos. Rapp. P. - V. Reun., Comm. Inst. Explor. Sci. Mer. Mediterr., Monaco. 22: 161-163.

Grant, B.R. 1971. Variation in silicate concentration at Port Hacking Station, Sydney, in relation to phytoplankton growth. Aust. J. Mar. Freshwater Res. 22: 49-54.

Hasler, A.D. 1974. Silica and nitrate depletion as related to rate of eutrophication in Lakes Michigan, Huron, and Superior. In: Coupling of land and water systems. SpringerVerlag, Berlin (GFR) London (UK).

Johnson, M., and W.D. Johnson. 1972. Occurrence of silica in the natural waters of the Huntley-Robertson district, southern New South Wales. Aust. J. Mar. Freshwater Res. 23: 105-119.

Kido, K. 1974. Latitudinal distribution and origin of particulate silica in the surface water of the North Pacific. Mar. Chem. 2: 277-285.

Kilham, P. 1971. A hypothesis concerning silica and the freshwater planktonic diatoms. Limnol. Oceanogr. 16: 10-18.

Lannergren, C. 1975. Phosphate, silicate, nitrate, and ammonia in Lindaspollene, a Norwegian land-locked fjord. Sarsia 59: 53-66.

Lewis, W.M., and F.H. Weibezahn. 1976. Chemical energy flow and community structure in some Venezuelan freshwater. Arch. Hydrobiol. (Suppl.) 50: 145-207.

Mann, C.R., A.R. Coote, and D.M. Garner. 1973. The meridional distribution of silicate in the western Atlantic Ocean. Deep Sea Res. Oceangr. Abst. 20: 791-801.

Morelock, J. 1970. Silica in the marine environment. Lagena (no. 25-26): 17-21.

Paasche, E. 1975. Growth of the plankton diatom Thalassiosira nordeaskioeidii Cleve at low silicate concentrations. J. Exp. Mar. Biol. Ecol. 18: 173-183.

Parker, B.C. 1969. Occurrence of silica in brown and green algae. Can. J. Bot. 47: 537-540.

_____ 1968. Occurrence of silica in brown algae. J. Phycol. (Suppl.) 4: 8.

Stephans, C.F., and C.H. Oppenheimer. 1972. Silica contents in the Northwestern Florida Gulf Coast. Contrib. Mar. Sci., Univ. Texas 16: 99-108.

Titman, D. 1976. Ecological competition between algae. Science 192: 463-465.